

IN THE CLAIMS:

Please add the following claims.

117. A purified and isolated nucleic acid sequence encoding a glial cell line-derived neurotrophic factor, wherein said nucleic acid sequence:

- (a) comprises nucleotides 304 through 705 of SEQ ID NO:3 or nucleotides 105 through 506 of SEQ ID NO:5;
- (b) encodes a polypeptide comprising an amino acid sequence set forth in SEQ ID NO:4 or SEQ ID NO:6;
- (c) encodes a polypeptide comprising an amino acid sequence which is in excess of 70% identical to an amino acid sequence set forth in SEQ ID NO:4 or SEQ ID NO:6 when up to four gaps in a length of 100 amino acids may be introduced to assist in that alignment;
- (d) hybridizes under conditions of reduced stringency to a nucleic acid sequence complementary to those defined in (a), (b) or (c), wherein said conditions include hybridizing sequences in 6X SSPE and 0.1% SDS at 42 to 50°C followed by washing in 2X SSPE and 0.1% SDS at room temperature to 50°C; or
- (e) encodes a polypeptide encoded by a nucleic acid sequence defined in (a), (b), (c) or (d) but differs in codon sequence due to the degeneracy of the genetic code;

wherein said encoded glial cell line-derived neurotrophic factor has the capability to promote dopamine uptake in dopaminergic neurons.

118. A purified and isolated nucleic acid sequence encoding a glial cell line-derived neurotrophic factor, said nucleic acid sequence comprising nucleotides 105 through 506 of SEQ ID NO:5, wherein said factor has the capability to promote dopamine uptake in dopaminergic neurons.

119. A purified and isolated nucleic acid sequence encoding a glial cell line-derived neurotrophic factor comprising an amino acid sequence as set forth in SEQ ID NO:6, wherein said factor has the capability to promote dopamine uptake in dopaminergic neurons.

120. A purified and isolated nucleic acid sequence encoding a glial cell line-derived neurotrophic factor comprising an amino acid sequence which is in excess of 70% identical to an amino acid sequence of SEQ ID NO:4 or SEQ ID NO:6 when up to four gaps in a length of

100 amino acids may be introduced to assist in that alignment, and wherein said factor has the capability to promote dopamine uptake in dopaminergic neurons.

121. A purified and isolated nucleic acid sequence encoding a glial cell line-derived neurotrophic factor, comprising a sequence which hybridizes under conditions of reduced stringency to a nucleic acid sequence complementary to those encoding an amino acid sequence of SEQ ID NO:4 or SEQ ID NO:6, wherein said conditions include hybridizing sequences in 6X SSPE and 0.1% SDS at 42 to 50°C followed by washing in 2X SSPE and 0.1% SDS at room temperature to 50°C, and wherein said factor has the capability to promote dopamine uptake in dopaminergic neurons.

122. A purified and isolated nucleic acid sequence according to claim 117, 118, 119, 120 or 121 further comprising a codon encoding an amino-terminal methionine residue.

123. A purified and isolated nucleic acid sequence comprising a sequence complementary to a nucleic acid sequence according to claim 117, 118, 119, 120 or 121.

124. A purified and isolated nucleic acid sequence according to claim 118 or 119 further comprising nucleotides encoding a pre-pro amino acid sequence as set forth in SEQ ID NO:25 amino acid residues 1 through 77.

125. A purified and isolated nucleic acid sequence encoding a glial cell line-derived neurotrophic factor, wherein said nucleic acid sequence:

- (a) encodes a polypeptide comprising the pre-pro form of human glial cell line-derived neurotrophic factor as set forth in SEQ ID NO:25 amino acid residues 1 through 211;
- (b) encodes a polypeptide comprising an amino acid sequence which is in excess of 70% identical to an amino acid sequence set forth in SEQ ID NO:25 when up to four gaps in a length of 100 amino acids may be introduced to assist in that alignment;
- (c) hybridizes under conditions of reduced stringency to a nucleic acid sequence complementary to those defined in (a), wherein said conditions include hybridizing sequences in 6X SSPE and 0.1% SDS at 42 to 50°C followed by washing in 2X SSPE and 0.1% SDS at room temperature to 50°C; or
- (d) encodes a polypeptide encoded by a nucleic acid sequence defined in (a), (b) or (c) but differs in codon sequence due to the degeneracy of the genetic code;

wherein said encoded glial cell line-derived neurotrophic factor has the capability to promote dopamine uptake in dopaminergic neurons.

126. A vector comprising a nucleic acid sequence according to claim 117, 118, 119, 120 or 121.

127. A host cell transformed or transfected with a nucleic acid sequence according to claim 117, 118, 119, 120 or 121.

128. A host cell according to claim 127 wherein said host cell is an animal cell.

129. A host cell according to claim 127 wherein said host cell is a COS cell.

130. A host cell according to claim 127 wherein said host cell is a microorganism.

131. A host cell according to claim 127 wherein said host cell is a bacterial cell.

132. A host cell according to claim 127 wherein said host cell is an *E. coli* cell.

133. A host cell according to claim 127 wherein said cell is transformed or transfected *ex vivo* and wherein said cell expresses and secretes said glial cell line-derived neurotrophic factor.

134. A host cell according to claim 127 wherein said cell is suitable for human implantation and wherein said cell expresses and secretes said glial cell line-derived neurotrophic factor.

135. A host cell recombinantly transformed or transfected to express a nucleic acid sequence encoding a glial cell line-derived neurotrophic factor, wherein said nucleic acid sequence:

- (a) comprises nucleotides 105 through 506 of SEQ ID NO:5;
- (b) encodes a polypeptide comprising an amino acid sequence set forth in SEQ ID NO:4 or SEQ ID NO:6; or
- (c) encodes a polypeptide comprising an amino acid sequence which is in excess of 70% identical to an amino acid sequence set forth in SEQ ID NO:4 or SEQ ID NO:6 when up to four gaps in a length of 100 amino acids may be introduced to assist in that alignment;

wherein said encoded glial cell line-derived neurotrophic factor has the capability to promote dopamine uptake in dopaminergic neurons

136. A purified and isolated nucleic acid sequence encoding a glial cell line-derived neurotrophic factor polypeptide comprising an amino acid sequence which is at least 90% identical to an amino acid sequence of SEQ ID NO:4 or SEQ ID NO:6 when up to four gaps in a length of 100 amino acids may be introduced to assist in that alignment and wherein said polypeptide has the capability to promote dopamine uptake in dopaminergic neurons.

137. A vector comprising a nucleic acid sequence according to claim 136.

138. A host cell transformed or transfected with a nucleic acid sequence according to claim 136.

139. A host cell according to claim 138 wherein said host cell is a microorganism.

140. A host cell according to claim 138 wherein said host cell is an animal cell.

141. A host cell according to claim 138 wherein said cell is suitable for human implantation and wherein said cell expresses and secretes said glial cell line-derived neurotrophic factor.

142. A host cell according to claim 138 wherein said cell is transformed or transfected *ex vivo* and wherein said cell expresses and secretes said glial cell line-derived neurotrophic factor.

143. A method for the production of glial cell line-derived neurotrophic factor, comprising the steps of:

- (a) culturing a host cell according to claim 127 under conditions suitable for the expression of glial cell line-derived neurotrophic factor; and
- (b) optionally, isolating said glial cell line-derived neurotrophic factor therefrom.

144. A method according to claim 143 wherein said host cell is an animal cell.

145. A method according to claim 144 wherein said host cell is a COS cell.

146. A method according to claim 143 wherein said host cell is a microorganism.
147. A method according to claim 146 wherein said host cell is *E. coli*.
148. A method according to claim 143 further comprising the step of refolding said glial cell line-derived neurotrophic factor.
149. A method for the production of glial cell line-derived neurotrophic factor, comprising the steps of:
  - (a) culturing a host cell transformed or transfected with a vector according to claim 126 under conditions suitable for the expression of glial cell line-derived neurotrophic factor; and
  - (b) optionally, isolating said glial cell line-derived neurotrophic factor expressed by said host cell.
150. A method for the production of glial cell line-derived neurotrophic factor, comprising the steps of:
  - (a) culturing a host cell containing a nucleic acid sequence encoding glial cell line-derived neurotrophic factor under conditions suitable for the expression of said factor, wherein said nucleic acid sequence is selected from the group consisting of:
    - (i) nucleotides 105 through 506 of SEQ ID NO:5;
    - (ii) nucleotides encoding a polypeptide comprising an amino acid sequence set forth in SEQ ID NO:4 or SEQ ID NO:6; or
    - (iii) nucleotides encoding a polypeptide comprising an amino acid sequence which is in excess of 70% identical to an amino acid sequence set forth in SEQ ID NO:4 or SEQ ID NO:6 when four gaps in a length of 100 amino acids may be introduced to assist in that alignment; and
  - (b) isolating said expressed glial cell line-derived neurotrophic factor in a substantially purified form from said host cell culture,  
wherein said glial cell line-derived neurotrophic factor has the capability to promote dopamine uptake in dopaminergic neurons.
151. A method according to claim 150 wherein said nucleic acid sequence further comprises a codon encoding an amino-terminal methionine.

152. A method according to claim 150 wherein said nucleic acid sequence encodes a factor comprising an amino acid sequence set forth in SEQ ID NO:4 or SEQ ID NO:6.

153. A method according to claim 150 wherein said nucleic acid sequence encodes a factor comprising an amino acid sequence which is in excess of 70% identical to an amino acid sequence of SEQ ID NO:4 or SEQ ID NO:6 when up to four gaps in a length of 100 amino acids may be introduced to assist in that alignment.

154. A method according to claim 150 wherein said host cell is an animal cell or microorganism.

155. A method according to claim 150 further comprising refolding expressed glial cell line-derived neurotrophic factor to form a disulfide-bonded dimer.

156. A method according to claim 150 wherein said glial cell line-derived neurotrophic factor is expressed by a bacterial host cell and is refolded to form a disulfide-bonded dimer.

157. A method according to claim 150 wherein the expressed glial cell line-derived neurotrophic factor is secreted by said host cell.

158. A purified and isolated nucleic acid molecule according to claim 117 which encodes glial cell line-derived neurotrophic factor having a molecular weight of about 31-42 kD on non-reducing SDS-PAGE, a molecular weight of about 20-23 kD on reducing SDS-PAGE, and which promotes dopamine uptake in dopaminergic neurons at a concentration of approximately 60 pg/ml.

Please cancel claims 26, 28, 29, 31, 34, 42-55, 75-86 and 95-116 without prejudice.

#### REMARKS

Claims 26, 28, 29, 31, 34, 42-55, 75-86 and 88-116 are pending in the application. Claims 88-94 have been allowed. The Examiner indicated that claims 28 and 34 were objected to, but no reasoning was provided in the office action. Assuming that the objection was that the claims were dependent upon rejected claim 26, the claims have been revised (new claims 118 and 119) and placed in independent form. The remainder of the previous claims have been